

Original Research Article

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Effect of Plant Extracts on Root-Knot Nematode *Meloidogyne incognita* Infecting Tomato

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ABSTRACT

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The root-knot nematode, *Meloidogyne incognita* is a key pest on tomato cultivation throughout the world. Eco-friendly technique for the management of *M. incognita* on tomato is still lacking. Hence, studies were conducted to test the nematicidal potency of leaf extracts of asparagus, white hibiscus, jasmine and Ixora plants against *M. incognita* under *in vitro* and pot culture. Under *in vitro* condition juvenile mortality rate was higher (49.4-90.2%) in asparagus treatment where as mortality rate was lower in Ixora treatment (8.2-35.4%). Pot culture experiment on the efficacy of plant extract on *M. incognita* revealed that the infection of root-knot nematode in terms of number of galls/plant was lower in asparagus leaf extract treatment. In other treatments, the number of galls varied from 33-57 / plant where as in control 100 galls/plant. Asparagus treated plants were taller with long root system. It is concluded that leaf extracts of asparagus is having high nematotoxic potential against *M. incognita* which can be recommended to tomato growers for root-knot nematode control after testing under field situations.

Introduction

Tomato (*Lycopersicon esculentum* L) is the world's most important vegetable crop that plays important role to meet nutritional requirement of people throughout the world. The fruits of tomato are also used in the preparation of soup, salad, pickles, ketchup, puree and sauces. Ripe tomato fruit has high nutritive value being a good source of vitamin A, B and C, potassium and minerals. Root-knot nematode *Meloidogyne incognita* is a plant-parasitic nematode that causes serious

damage on tomato in tropical and subtropical agriculture. Affected plants are often dwarfed, with small leaves. The infected tomato plants also express day wilt symptoms i.e. wilted appearance during day time and normal appearance during night time. The root-knot infected tomato express symptoms similar to nutrient deficiency. Tomato infested by *M. incognita* can lead to yield loss up to 80% (Nagachandrabose and Baidoo, 2017).

Management of nematodes in tomato can be achieved chiefly by chemical method.

However, repeated application of chemical nematicides is needed to get desirable nematode control. Continuous application of chemical nematicide can lead to pollution problem and toxicity to non-target organism (Seenivasan, 2017).

In the recent years many bio-control agents were tested against plant parasitic nematodes including *M. incognita* on tomato. However non-availability of their commercial formulations leads to restrict their usage by farmers. Hence, the requirement of the day for *M. incognita* control in tomato should be eco-friendly and locally available. Among the various eco-friendly strategies available for nematode management, application of extracts of locally available plants is one of the thrust idea need to be exploited for field level application (Seenivasan, 2011).

The concept of plant origin bio-pesticides in modern agriculture has now attained wider applications than earlier. A large number of plants/plant parts or their products have been tested in-vitro for their nematicidal attributes revealed that extractives of several plants prepared in water or in organic solvents have been found effective against several economically important plant parasitic nematodes (Thoden and Korthals, 2011). However, little information is available on the presence of the bioactive products present in Asparagus, White hibiscus, Jasmine and Ixora effective against plant-parasitic nematodes. Hence, this study was conducted with the following objectives viz.,

- 1) To study the direct mortality of *M. incognita* when exposed to leaf extracts of Asparagus, White hibiscus, Jasmine, Ixora,
- and 2) To study the effect of amending the soil with leaf extracts of Asparagus, White hibiscus, Jasmine and Ixora on *M. incognita* infection on tomato roots under pot conditions.

Materials and Methods

Culture of *Meloidogyne incognita*

A population of *Meloidogyne incognita* used for *in vitro* and glass house studies, was originally isolated from the tomato roots at Fruit Orchard, Agricultural College and Research Institute, Tamil Nadu Agricultural University, Madurai, Tamil Nadu, India. A single egg mass of *M. incognita* was collected from the infected tomato roots, placed in a Petri dish containing sterile water and allowed to hatch for a week. The juveniles hatched out were multiplied on tomato cv. Co 1 grown in pots (5 kg capacity) containing sterile red earth: sand: farmyard manure (2:2:1) mixture in the glasshouse. After 8-10 weeks, eggs and juveniles were collected from the infected tomato roots by a modified NaOCl extraction method (Hussey and Barker, 1973; Seenivasan and Senthilnathan, 2018). The entire root system was dipped in water and soil was removed gently without detaching egg sacs. Eggs were extracted by vigorous shaking of infested roots in a 1% sodium hypochlorite solution for 3 min. The resulting suspension was then passed through a range of deferent mesh-size sieves. The eggs were collected on a fine sieve (350 mesh) and washed in tap water to remove all traces of sodium hypochlorite before use. Hatched juveniles of *M. incognita* were obtained by placing the eggs in sterile distilled water for 5 days at room temperature. The freshly hatched juveniles (J2) were used for the laboratory and pot tests.

Preparation of leaf extracts

Five g of leaves was taken in a conical flask and 80 ml of methanol was added. It was kept in the shaker overnight at 40°C at 130 rpm, again 80 ml of methanol was added and kept in shaker. This process was repeated for five times to get leaf extract concentrate. The

prepared plant extracts were used for *in vitro* and pot tests.

Effect of plant extracts on second stage juveniles (J2) of *Meloidogyne incognita*

To determine the effect of various concentrations of plant extracts (25%, 50% and 100%) on mortality of *M. incognita* second stage juveniles (J2), 9 ml of each plant extract concentrations was poured into small petri dishes to which 1 ml suspension containing freshly hatched J2 containing 20 J2 ml⁻¹ was added. One ml of juvenile suspension in nine ml of sterile distilled water served as control. Each treatment was replicated five times. For J2 mortality test, the number of dead juveniles were counted after 48 h exposure and expressed as per cent mortality. Nematode was considered dead if they did not move when probed with a fine needle.

Evaluation of plant extracts against *M. incognita* in tomato (Pot culture experiment)

The plant extracts of Asparagus, White hibiscus, Jasmine and Ixora were tested against *M. incognita* infesting tomato under glasshouse conditions. Pot culture experiment was conducted at the Department of Plant Pathology, Agricultural College and Research Institute, Madurai during February 2019 to April 2019. Tomato seed PKM1 obtained from Horticultural College and Research Institute, Periyakulam was used in this experiment. One month old tomato seedlings from nursery pots were carefully uprooted, washed thoroughly to remove the soil particles and transplanted @ one/pot in 15 cm diameter earthen pots containing steam-sterilised pot mixture (Red soil: Sand: Farmyard manure; 3: 2: 1) and maintained in the glasshouse. At the time of sowing, the following treatments were applied to the soil

in each pot and mixed thoroughly; T1 – Asparagus extract; T2 - White *Hibiscus* extract; T3 – Jasmine extract; T4 – *Ixora* extract; and T5 - Untreated control. A completely randomized design was adopted with four replications for each treatment. Five days after sowing, freshly hatched, juveniles of *M. incognita* were inoculated in the root zone at 500 J2/pot. The plants were carefully uprooted after a month after inoculation and the observations on number of galls, shoot length and root length were made.

The data collected from both studies were analyzed and compared by *t*-test following Panse and Sukhatme (Panse and Sukhatme, 1954).

Results and Discussion

Efficacy of *Meloidogyne incognita* in different plant extracts under *in vitro* conditions

It was observed that all the tested concentrations of aqueous extracts (25%, 50% and 100% or crude) *viz.*, asparagus, white hibiscus, jasmine, Ixora caused mortality of *M. incognita* juveniles as compared with control. However, mortality rate significantly differed among tested plant extracts and even within their concentrations. Extract of asparagus was best among tested plant extracts which recorded 90.2% mortality while white hibiscus has mortality 74.3%, jasmine recorded mortality of 50.4% and ixora has mortality rate of 35.4% in crude concentration (Figure 1).

At 50% concentration of the asparagus extract, the maximum mortality of juveniles (79.1%) was recorded. It was followed by white hibiscus (48.4% mortality), jasmine (35.6% mortality) and Ixora (15.5% mortality). In 25% concentration, the extract of asparagus has juvenile mortality of 49.4%,

white hibiscus recorded mortality of 23.5%, jasmine recorded mortality of 14.7% and Ixora recorded 8.2% of mortality (Figure 1).

The results clearly evidenced that Asparagus leaf extract is the best for managing *M. incognita* than leaf extracts of white hibiscus, jasmine and ixora. The major bioactive constituents of Asparagus are a group of steroidal saponins. Other primary chemical constituents of Asparagus are essential oils, arginine, tyrosine, flavonoids and resin and tannin. The other bioactives principles are sarsasopogenin and shatavarin 1-4 which are present in leaves of Asparagus species. The nematicidal properties of root exudates of asparagus was first reported against the stubby root nematode, *Trichodorus christiei* [8]. They proved that a low molecular weight glycoside compound with aglycone is responsible *T. christiei* mortality. The asparagusic acid present in roots of asparagus was reported to have nematicidal property against several plant parasitic nematodes (Rohde and Jenkins, 1958). Earlier studies established that the compounds from asparagus root exudates and acid from asparagus root are responsible for nematode control. This work will be the first to establish the nematicidal properties of leaf extracts of asparagus against *M. incognita*. The presence

of asparagusic acid was reported in seeds of asparagus plants (Takasugi *et al.*, 1975). Results of our study also suggested that the leaf of asparagus might possess glycoside like compound or asparagusic acid that might be studied further to prove this hypothesis.

Effect of plant extracts on galling of roots of tomato inoculated with *Meloidogyne incognita* under pot culture condition

The results revealed that lesser number of galls was produced by *Meloidogyne incognita* in the plants treated with Asparagus extract which was significant over the other treatments and the control (Figure 2). It was also observed that infectivity of root-knot nematode *Meloidogyne incognita* was not inhibited much with Ixora extract treatment. Earlier the root extracts of asparagus was evaluated against *M. incognita* on egg plant and results are in confirmatory with our findings (Izuogu *et al.*, 2012). To our knowledge, this is the first report that evidenced the negative effect of leaf extracts of asparagus on *M. incognita* infectivity on tomato. The bioactive compounds present in the leaf extracts of asparagus caused mortality of *M. incognita* that affected the infectivity on tomato plants. The results obtained *in vitro* studies supports this hypothesis (Table 1).

Table.1 Effect of different plant extracts on growth of tomato challenge inoculated with *Meloidogyne incognita*

Treatments	Shoot length(cm)	% increase over control	Root length (cm)	% increase over control
ASPARAGUS	17.8	16.8	8.4	7.4
JASMINE	10.3	9.3	5.7	4.7
WHITE HIBISCUS	13.7	12.7	6.3	5.3
IXORA	8.7	7.7	4.3	3.3
CONTROL	6.9	-	4.1	-
CD at P=0.05	2.8	-	1.9	-

Fig.1 Mortality rate of root knot nematode *Meloidogyne incognita* when exposed to different plant extracts under *in-vitro* condition

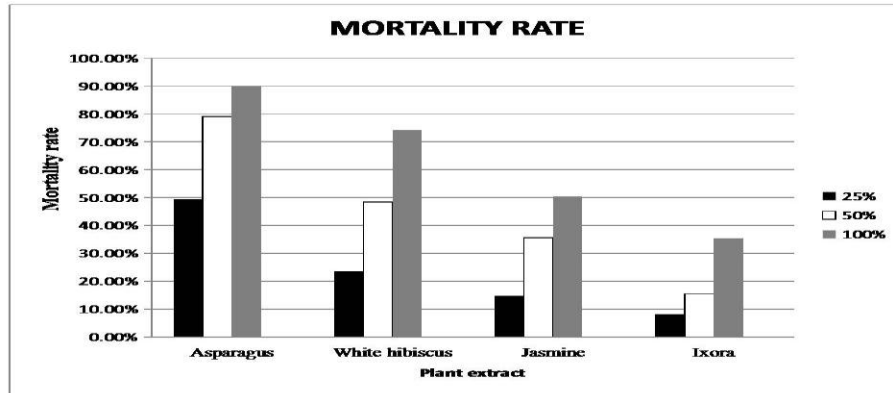


Fig.2 Infectivity of *Meloidogyne incognita* on tomato after treatment with plant extracts

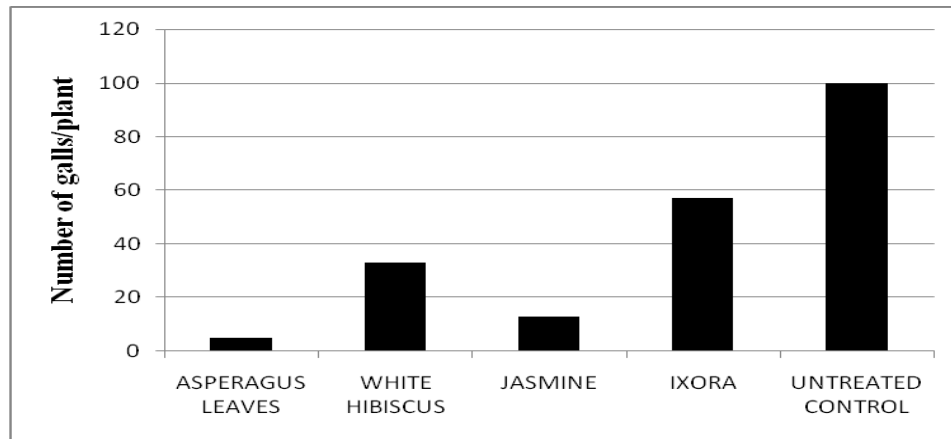


Fig.3 Gallling pattern in asparagus treated and untreated tomato plant roots



Untreated control



Asparagus extract treated plant

The plants treated with Asparagus registered the maximum shoot length (17.4 cm) and root length (8.7 cm) (Figure 3). The profound improvement in growth of tomato might be due to low *M. incognita* infectivity in asparagus treated plants. Similar results of improved growth due to *M. incognita* reduction were established earlier (Seenivasan, 2010).

From the findings of this research work, it can be recommended that soil application of methanol extracts of asparagus leaves can be followed by the tomato growers to manage root-knot nematode menace. It is a low cost technology i.e. the farmer itself can prepare asparagus leaf extracts as it is a common garden land weed plant. This technology can also become a replacement for the synthetic, dangerous and expensive chemical nematicide. However, further efficacy tests under field conditions are essential before adoption by farming community.

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